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THE EFFECTS OF DOUGLAS FIR MONOCULTURE ON STAND CHARACTERISTICS IN A ZONE OF MONTANE BEECH FOREST

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Abstract: The right choice of tree species to form forest cultures is of paramount importance to the preservation of the diversity, fertility and ecological stability of forest ecosystems. To that end, we examined the effect of a 40-year-long cultivation of Douglas fir (*Pseudotsuga menziesii* (Mirb) Franco) on the floristic composition, characteristics of the forest floor, physical and chemical properties of the soil and the intensity of organic matter decomposition in a beech forest in western Serbia (Mt. Maljen). It was found that the cultivation of Douglas fir caused a reduction in biodiversity, changes in the chemical properties of the soil, that were most pronounced in the surface layers (0-10 cm), and a slowing down in the metabolism of the beech stand. The absence of many plant species characteristic to natural beech forests was observed in the Douglas fir plantation, these were reflected in the detected changes in the chemical properties of the soil, such as lower substitutional acidity ($p<0.05$), depletion of the adsorption of basis in the cation complex ($p<0.001$) and lower amounts of C, N, P ($p<0.001$) and K ($p<0.01$) in relation to the beech stand (control). No differences were found in soil moisture and active acidity levels. The higher value of the C/N ratio of the Douglas fir litter ($p<0.001$) provided proof for its lower decomposition rate compared to beech litter ($p<0.05$). Over time, all these changes could lead to further acidification and degradation of the soil and a reduction in this ecosystem's productivity.

Key words: beech; Douglas fir monoculture; degraded habitat; soil characteristics; biodiversity

INTRODUCTION

Woody species have a crucial role in nutrient cycling and in the maintenance of fertility and ecological stability of forest ecosystems [1,2]. For this reason, the substitution of tree species can have considerable impact in the modification of the physical and chemical characteristics of soil and on the processes in the soil [3,4]. These changes are influenced by differences in the ability of woody species to enhance atmospheric deposition, which in turn affects the chemical properties of their litter and the formation of diverse microclimatic conditions. Nonetheless, in modern forest ecology and forest soil science, litter quality of the dominant woody species and its decomposition rate are the primary factors affecting key soil processes, particularly the nutrient cycles responsible for soil ecosystem functioning and stability. In general, due to the presence of recalcitrant components in their needles, coniferous species reduce the cycling of nutrients and microbial activity, enhancing the acidification of the soil [9]. The lower content of nutrients and cations in the litter of coniferous species leads to a reduction in base saturation (V%), cation exchange capacity (T), pH, soil fauna activity, decomposition rate, total porosity and widening C/N ratios [10,11,4].

The growing needs of human society for wood have shaped forest ecosystems throughout Europe, where, in order to achieve high productivity, large areas of mixed and deciduous forest stands are replaced by coniferous monocultures, most often spruce (*Picea abies* Karst.); thus the average composition of European forests has changed at the expense of their biodiversity [4,12-14]. The widespread acidification of soil and reduction in vitality of these forests throughout Europe show that monodominant spruce forests present a very high risk in terms of climate change, which has become especially pronounced in recent decades. It has been found that the cultivation of spruce at low altitudes (up to 900 m) often contributes to the development of dystric cambisols and podzols, i.e. acidic or very acidic soil, poor in nutrients [11,20,21]. In the conversion practice used in many central European countries to resolve these problems, Douglas fir (*Pseudotsuga menziesii* (Mirb) Franco) has acquired a prominent place thanks to its high adaptive capacity and the significant economic potential of its high production capacity. Today it represents the most important cultivated commercial non-native tree species in northern and central Europe, where it thrives in different habitats and creates productive forest stands and plantations [14,22]. However, the modern concept of multifunctional forestry not only entails the realization of high productivity and profit, but also the provision of important ecosystem functions, such as soil fertility, high quality water resources and nitrogen and carbon sequestration [23,24].

According to the data of the National Forest Inventory, in central Serbia the total area of artificial stands (cultures and plantations) is 174800 ha, or 7.8% of the total forest area, of which 71.4% are coniferous cultures. In the large-scale introduction of conifers in coppiced and degraded beech forests during the seventies, translocated autochthonous (spruce – *P. abies* and black pine – *Pinus nigra* J. F. Arnold) and allochthonous coniferous species (Douglas fir – *P. menziesii* and Weymouth pine – *Pinus strobus* L.) were used [25]. However, in central Serbia, stands of Douglas fir are only found in some localities, making up only 0.2% of the total wood volume. The small representation of Douglas fir in afforestation and reclamation of forestland in central Serbia continues today and is illustrated by the fact that in the afforestation carried out in 2008 and 2009, only 25 ha of Douglas fir were planted as opposed to the 1865 ha of planted spruce [26]. The reason for this low representation of Douglas fir in Serbian forests probably lies in the small number of provenance tests, which are essential when introducing species into new habitats species that in their native habitat have such a wide radius of natural distribution (between 19° and 55° N latitude) [14,27]. The successful introduction of Douglas fir and the establishing of viable cultures require long-term examination of all the characteristics of the species in order to select the most adaptive and most productive provenance for seed collection [28].

Worldwide, the study of Douglas fir mostly focuses on the analysis of different silvicultural treatments, as well as the determination and augmentation of its productive characteristics by the application of various fertilization treatments in order to optimize production and profit [13,29-32]. An ecological approach to research that examines its impact on the dynamics of the forest floor and on the diversity of the habitat at the time of introduction, especially compared to natural beech forests, is even less common [2,8,14,33-35]. Not enough is known of how these introduced species affect the soil and biogeochemical cycle of nutrients through soil horizons [36]. Examinations of the effects of substitution of spruce and black pine by pure and mixed stands of Douglas fir on soil characteristics have revealed that the influence of this exotic species on soil chemistry, organic matter and nutrient dynamics is more favorable than other coniferous species [24,34,37,38]. Namely, it has been shown that Douglas fir acidifies the upper soil layers to a lesser extent and contributes to the creation of more favorable forms of humus compared to spruce. However, from an ecological and conservation standpoint, the cultivation of the very invasive Douglas fir, particularly in pure and dense stands that cover large areas, can have seriously deleterious consequences on the forest ecosystem [14]. Recent studies have mostly focused on only one aspect of the effects of Douglas fir, examining its impact on either the soil characteristics [2,8] or the characteristics and diversity of understory species [35,39]. However, there is a lack of research including both interactions. For this reason, the aim of this work was to determine whether the forty-year-long cultivation of Douglas fir in a montane belt of beech forest in western Serbia affected the floristic composition, the physical and chemical soil characteristics, and the metabolism of this habitat. Our research into the effects of Douglas fir cultivation included both aspects: the causal relationship between Douglas fir and the soil, and between the soil and the understory. The results of this study contribute to a better understanding of the negative effects of logging and substitution of deciduous with conifer tree species in forest ecosystems, where the sustained provision of fertile soil, carbon sequestration and nitrogen retention are of invaluable importance to their long-term stability.

MATERIALS AND METHODS

Study site

The study site is located in western Serbia, on Mt. Maljen (lat/lon 44°10'N/20°5'E), in the locality of Kaona (880 m a.s.l.), in a climatoregional area of montane beech forests (*Fagetum montanum* s. lat). The climatic conditions of this area are moderate continental (mean annual temperature 9.16°C, average temperature during the vegetation period 15°C, and mean annual precipitation 890 mm). According to Lang's index of climate types, the climate on Kaona is semi-humid (rain factor RF=98.2), while Kerner's thermodromic coefficient (K=9.52) shows that this climate is submaritime.

Experimental areas

Forty years ago, in a beech forest of coppice origin, the clear-cutting of beech trees was carried out in 20 m-wide strips. Douglas fir stands were established in these clearings, with these strips alternating with areas populated by beech. Our experimental sites were a Douglas fir stand strip and a strip of autochthonous beech forest (control). The criterion for their selection was that they were situated on the same soil type (dystric cambisol), formed on the same bedrock (diabase), with the same exposure (west) and almost the same terrain incline (2-5°).

Floristic composition

Phytocoenological research was conducted according to the Westhoff and Van der Marrel method [40] in 20x20 m plots. For each area, three phytocoenological releves were taken. The plant species were determined based on the Phytosociological Analysis of Forest Vegetation of Maljen [41] and Iconographia Florae Partis Austro-Orientalis Europae Centralis [42].

Litter and soil analysis

For the litter samples (OLF horizon-layer, n=7), dried at 65°C to a constant weight and ground to pass through a 0.5 mm sieve, the pH was determined in deionized water (0.6 g of plant material/15 ml H_2O) and 1 N KCl (0.6 g of plant material/15 ml 1 N KCl + 1.2 g $BaSO_4$). The total carbon and nitrogen content in the litter was determined from the same sample using the modified Anstet method [43].

Soil moisture was determined in the period from April to October at 10-cm increments up to a depth of 80 cm, with five replications. Moisture levels were established gravimetrically, using a Chyo IB 30 type hygrometer. The results are presented as the average levels of soil moisture during the vegetation period. To establish the average amount of litter, litter samples of the studied species were collected in the same time interval (from April to October), using a metal square with an area of 50 x 50 cm with five replications, dried at 105°C, measured and expressed per 1 ha.

The chemical properties of the soil were determined at soil depths of 0-10 cm, 10-20 cm, 20-50 cm and 50-80 cm with 7 replications (n=7). The samples were pre-dried at 65°C to a constant weight and sieved. The acidity of the soil (active and substitutional) was determined potentiometrically, with a glass electrode, using a mixture of soil/deionized water and soil/1N KCl (1:2.5, w/v). Soil adsorptive complex characteristics were determined according to Kappen: the content of exchangeable bases (S, cmol kg^{-1}), hydrolytical acidity, the sum of acidic cations (T-S, cmol kg^{-1}), and cation exchange capacity (T, cmol kg^{-1}) were calculated. The saturation of the adsorption complex with bases (V%) was determined according to Hissink. The total organic carbon content (C, g kg^{-1}) in the soil was determined by potassium dichromate oxidation using Simakov's modification of the Turin method [44], and the total nitrogen content (N, g kg^{-1}) using the semimicro-Kjeldahl method. The C/N ratio was calculated. Available phosphorus (P_2O_5, g kg^{-1}) and potassium (K_2O, g kg^{-1}) were extracted with ammonium acetate-lactate (A-L solution, pH 3.7, ratio 1:20) and determined by flame photometry [45].

Litter decomposition

In order to determine the rate of decomposition of the leaf litter samples, the 'litterbag method' was used [46-48]. The decomposition rate for the organic matter of beech and Douglas fir was determined 6 and 12 months after the start of decomposition. Freshly fallen leaves and needles from the examined species were collected on plastic nets spread out on the surface of the forest soil. Leaf litter (20 g), dried at a temperature of 65°C to a constant weight, was enclosed in a bag (20x20 cm) made of plastic netting with a mesh size of 1 mm. In October, ten bags were randomly placed for each species on the surface of the forest floor. After six months, in April of the following year, five of the bags were sampled for each species (n=5), and after twelve months (October) the remaining five bags were collected (n=5) from both plots. Once the soil and roots were removed, all the bags were dried at 65°C to a constant weight in order to determine the remaining weight of the organic matter.

Based on Olson's decomposition model and Olson's rate constant of loss [49], the prognosis for the decomposition of organic matter from beech and Douglas fir was calculated using the formula:

$$\frac{\mathrm{Mt}}{Mo} = \mathrm{e}^{-kt}$$

where M_0 is the initial mass of the organic matter, M_t is the mass of the organic matter after a year (t) of decomposition and k is Olson's rate constant of loss after 12 months of decomposition; $k_{1/2}$ represents the decomposition coefficient after 6 months. According to this exponential model, half of the decomposition time, $t_{1/2}$=0.639*k; the time constant, k=0.368 of 1/e. The time required (in years) for the decomposition of 95% of the organic matter was calculated using 3/k, while the time required (in years) for the decomposition of 99% of the organic matter was calculated using 5/k.

Statistical analysis

One-way analyses of variance (ANOVA) were performed to test the differences between the Douglas fir stand and the beech stand, which served as the control, in terms of the physical and chemical characteristics of the soil, as well as the litter characteristics. Soil moisture, soil and litter acidity (active and potential), characteristics of the adsorptive complex of the soil (cation exchange capacity (T), content of exchangeable bases (S), sum of acidic cations (T-S), saturation of the adsorption complex with bases (V), total nitrogen content, available P and K in soil, as well as the C/N ratio in soil and litter, were compared. The intensity of the decomposition of Douglas fir and beech organic matter after six and twelve months was also compared, as were the decomposition constants ($k_{1/2}$ and k) and decomposition prognosis constants (3/k and 5/k) for the organic matter from these species (subsequent tests of normality using the Shapiro-Wilk W test and Levene's test of homogeneity of variances showed non-significant values for all the reported ANOVA breakdowns).

RESULTS

Floristic composition

The vertical distribution of the Douglas fir culture is separated into a tree layer with complete dominance of Douglas fir (*P. menziesii*, abundance 8-9, cover 75-100%) and in a herbaceous layer, while no shrub layer is present (Table 1). The Douglas fir has an average height of 15-18 m and trunk diameter of 30 cm. In this layer, thinning cuts were of weak intensity. In the herbaceous layer covering 3% of the area, *Rubus hirtus* W et K. (2-3), *Cardamine bulbifera* (L.) Crantz. (2-3), *Viola silvestris* Lam. (2), *Galeobdolon luteum* Hudson (2), *Fagus moesiaca* (Domin, Maly) Czeczott (2), *Pteridium aquilinum* (L.) Kuhn. (2), *Sambucus nigra* L. (2) and *Galium odoratum* (L.) Scop. (2) are found. In the control beech stand, three layers are separated. In the tree layer there is only beech (*Fagus moesiaca* (Domin, Maly) Czeczott, abundance 7-9, cover 50-100%). The trees are approximately 80 years old with an average height of 18-20m and trunk diameter of 35-40cm. The shrub layer covers 10% of the surface area, while the layer of herbaceous plants covers 100% (Table 1).

Litter and soil analysis

The average values of soil moisture during the vegetation period in both examined areas decreased with the depth of the horizon. At both stands, the moisture content in the examined soil depths was similar, except at the depth of 50-80 cm. At this depth, the soil was found to be moister in the Douglas fir stand than in the control site (21.10±1.69%:18.10±2.62%, $p<0.01$) (Fig. 1).

Levels of active acidity of the soil, both in the litter layer and in all the investigated soil layers, were higher than those of substitutional acidity (Table 2).

Table 1. Analysis of the floristic composition in beech stand (control) and Douglas fir (plantation).

Community	***Fagetum montanum* - control**			***Pseudotsuga menziesii* - plantation**		
Releve number	1	2	3	4	5	6
Locality	Maljen - NW Serbia (Kaona)					
Altitude (m)	880					
Exposition	W					
Slope (°)	5	5	5	2	2	2
Bedrock	Diabase-chert formation					
Soil	Distrc cambisol					
Average dbh (cm)	35	40	35	30	30	30
Average height (m)	18	20	18	15	18	18
Size of sample area (m)	20x20					
Tree layer						
Fagus moesiaca (Domin, Maly) Czeczott	7	9	8			
Pseudotsuga menziesii				8	9	8
Shrub layer						
Fagus moesiaca (Domin, Maly) Czeczott	3	3	3			
Carpinus betulus L.	2		3			
Crataegus monogyna Jacq.	2		3			
Corylus avellana L.	2		3			
Acer pseudoplatanus L.	3					
Rubus hirtus W. et K.		2				
Prunus avium L.						
Cornus mas L.		2				
Betula pendula Roth		2				
Sambucus nigra L.		2				
Salix caprea L.		2				
Herb layer						
Rubus hirtus W et K.	8	7	7	3	2	2
Cardamine bulbifera (L.) Crantz.	3	3	3	2	3	2
Viola silvestris Lam.	3	3	3	2	2	2
Galeobdolon luteum Hudson	2	3	3	2	2	2
Fagus moesiaca (Domin, Maly)Czeczott	3	5	5	2	2	2
Pteridium aquilinum (L.) Kuhn.	2	2	5	2	2	2
Mycelis muralis (L.) Dum	3	2	3			
Sambucus nigra L.		2				2
Galium odoratum (L.) Scop.	5	5	3		2	2
Asperula taurina L.	3	2				
Acer pseudoplatanus L.	2	2	2			
Allium ursinum L.	3	2	3			
Helleborus odorus W. et K.	2	2	2			
Epilobium montanum L.		2	2			
Stachys silvatica L.	2					
Crataegus monogyna Jacq.			2			
Quercus cerris L.			2			
Circaea lutetiana L.	2					
Clinopodium vulgare L.	2		3			
Moehringia trinervia (L.) Clariv.	2		3			
Glechoma hirsuta W. et K.	3		3			
Carpinus betulus L.	2		2			
Corylus avellana L.	2		2			
Sanicula europaea L.	3		2			
Galium silvaticum L.			2			
Carex silvatica Hudson			2			
Euphorbia amygdaloides L.			3			
Aremonia agrimonoides (L.) Neck.			2			
Polygonatum multiflorum (L.) All.	2		2			
Symphytum tuberosum L.	2		2			
Scrophularia nodosa L.	2		2			
Veronica chamaedrys L.		2	2			
Clematis vitalba L.			2			
Urtica dioica L.			2			

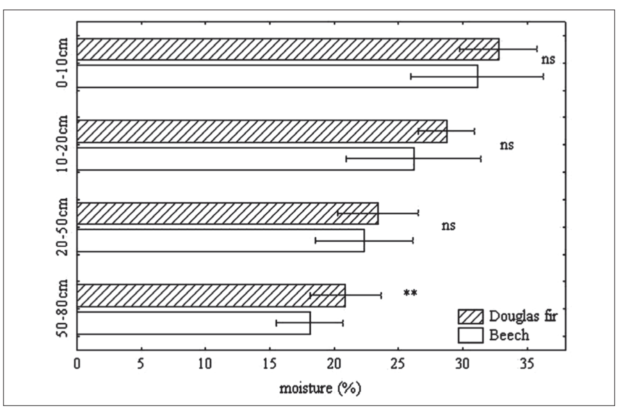

Fig. 1. Soil humidity in a beech forest and a Douglas fir plantation (ANOVA, n=5, level of significance: **p<0.01, ns – not significant).

Statistically significant differences were noted only for substitutional acidity at a depth of 0-10 cm, with lower values found in the Douglas fir stand than in the control site (substitutional acidity 3.36±0.27:4.01±0.32, $p<0.05$). In the litter layer, as well as in the deeper layers of soil, no differences between the sites were noted in terms of active and substitutional acidity (Table 2).

Cation exchange capacity (T cmol kg^{-1}) was lower in the Douglas fir stand than in the control site at soil depths of 0-10 cm (44.81±3.15:62.75±4.26, $p<0.001$), 10-20 cm (39.66±3.51:51.18±4.86, $p<0.001$) and 20-50 cm (21.72±1.54:24.95±1.69, $p<0.01$), and higher at a soil depth of 50-80 cm (28.71±3.12:21.16±2.31

Table 2. Chemical properties of soil and litter in a beech stand (control) and Douglas fir plantation.

Horizon	Tree spec.	pH				Adsorptive complex								C		N		C/N	
						T		S		T-S		V							
		H_2O		KCl		cmol kg^{-1}						%		g kg^{-1}				%	
OLF	Df	4.95±0.36	ns	4.24±0.28	ns	-		-		-		-		335.9±34.8	ns	12.1±0.9	***	27.76±2.31	***
	B	5.06±0.48		4.53±0.32		-		-		-		-		375.4±37.7		17.8±1.5		21.09±1.98	
0-10cm	Df	4.70±0.28	ns	3.66±0.27	*	44.81±3.15	***	10.23±1.15	***	34.58±2.95	***	22.83±1.76	***	63.4±11.6	***	4.5±0.5	***	14.09±1.22	***
	B	4.86±0.35		4.01±0.32		62.75±4.26		20.66±1.98		42.09±3.37		32.92±2.95		104.0±14.5		8.8±1.0		11.82±0.96	
10-20cm	Df	4.77±0.38	ns	3.69±0.25	ns	39.66±3.51	***	8.66±0.78	***	31.00±2.94	***	21.84±1.83	ns	53.0±8.7	**	4.4±0.5	***	12.04±1.23	**
	B	4.79±0.41		3.70±0.28		51.18±4.86		11.21±1.02		39.97±3.14		21.90±1.99		73.0±11.6		7.1±1.1		10.28±1.01	
20–50cm	Df	5.27±0.51	ns	3.88±0.31	ns	21.72±1.54	**	5.90±0.49	***	15.82±1.13	***	27.10±2.45	***	14.6±2.9	**	2.0±0.2	ns	7.41±0.80	***
	B	5.27±0.46		3.94±0.33		24.95±1.69		4.72±0.45		20.23±1.86		18.92±1.33		20.1±4.1		2.1±0.3		9.62±0.79	
50–80cm	Df	5.48±0.55	ns	3.82±0.40	ns	28.71±3.12	***	17.61±2.04	***	11.10±1.34	ns	61.34±5.59	***	5.4±1.7	ns	0.8±0.0	ns	6.75±0.73	**
	B	5.54±0.52		3.90±0.45		21.16±2.31		9.25±1.31		11.91±1.32		43.71±3.98		8.1±2.9		1.0±0.2		8.10±0.75	

ANOVA, n=7, Df – Douglas fir, B – beech, level of significance: ***p<0.001, **p<0.01, *p<0.05, ns-not significant

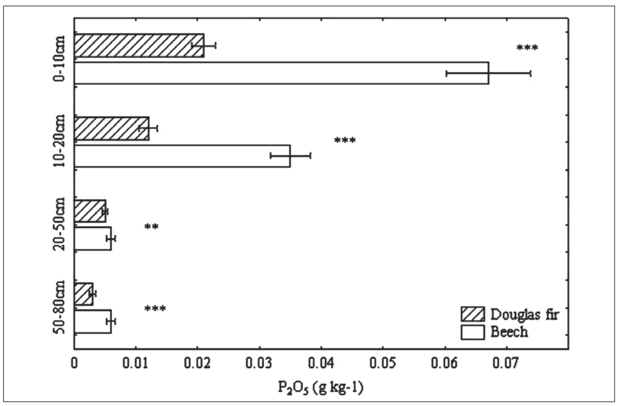

Fig. 2. Concentration of available phosphorus (P_2O_5) in the soils of a beech forest and a Douglas fir plantation (ANOVA, n=7, level of significance: ***p<0.001, **p<0.01).

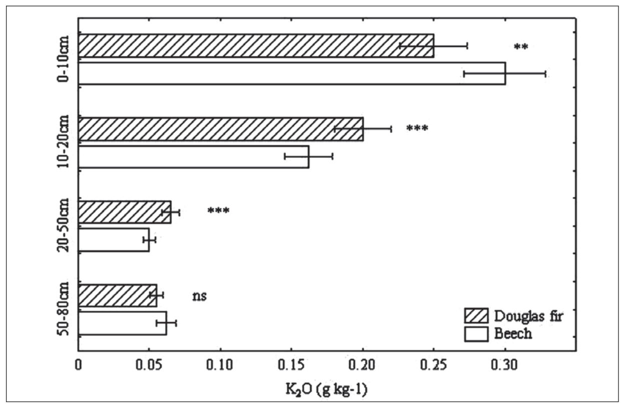

Fig. 3. Concentration of available potassium (K_2O) in the soils of a beech forest and a Douglas fir plantation (ANOVA, n=7, level of significance: ***p<0.001, **p<0.01, ns - not significant).

p<0.001) (Table 2). Lower levels of the content of exchangeable bases (S cmol kg^{-1}) were recorded in the surface layer of soil (0-10 cm) in the Douglas fir stand compared to the control site (10.23±1.15:20.66±1.98, p<0.001), and at depths of 10-20 cm (p<0.001) and 20-30 cm (p<0.001), while at a soil depth of 50-80 cm, the values of S were higher in the Douglas fir stand than in the control site (17.61±2.04:9.25±1.31, p<0.001) (Table 2). The sum of acidic cations (T-S cmol kg^{-1}) in the Douglas fir culture soil was lower than that in the control stand at depths of 0-10 cm (34.58±2.95:42.09±3.37, p<0.001), 10-20 cm (31.00±2.94:39.97±3.14, p<0.001) and 20-50 cm (15.82±1.13:20.23±1.86, p<0.001), while at the depth of 50-80 cm differences were not noted (Table 2). In the surface layer of soil (0-10cm), lower levels in the degree of saturation of the adsorption complex with bases (V%) were noted in the Douglas fir stand (22.83±1.76:32.92±2.95, p<0.001) (Table 2). As the depth increased, the degree of saturation of the adsorption complex with bases in the Douglas fir stand increased, and in the deeper layers of soil (50-80 cm), it became greater than at the control beech stand (61.34±5.59:43.71±3.98, p<0.001).

The litter of both examined species had a similar carbon content (C g kg^{-1}), while the nitrogen content (N g kg^{-1}) was greater in beech litter than in Douglas fir litter (17.80±1.10:12.10±0.90, p<0.001) (Table 2). Analysis of essential nutrients, total N (N g kg^{-1}) and easily accessible P (P g kg^{-1}) and K (K g kg^{-1}) in the soil showed that their content declined with horizon depth at both sites (Table 2, Figs. 2, 3). The N content was lower in the Douglas fir culture soil at depths of 0-10 cm (4.50±0.50:8.80±1.00, p<0.001) and 10-20 cm (4.40±0.50:7.10±1.10, p<0.001), while the content of easily accessible P was lower at all depths. The greatest differences were seen in the surface layers of soil at

Table 3. Amount of litter and intensity of organic matter decomposition in beech and Douglas fir stands.

Tree species	Amount of litter t ha^{-1}	F coef.	6 months M±SD (%)	F coef.	12 months M±SD (%)	F coef.
Beech	7.482 ± 2.368	2.882 **	18.2 ± 0.7	136.661 ***	32.76 ± 3.57	8.578 *
Douglas fir	12.791 ± 4.021		9.48 ± 1.51		24.98 ± 4.74	

ANOVA, n=5, values are mean (S.D.), levels of significance: ***p<0.001, **p<0.01, *p<0.05

Table 4. Analysis of the decomposition constants ($k_{1/2}$ and k) and prognosis constants (3/k and 5/k) of the organic matter in beech and Douglas fir stands.

Tree species	$k_{½}$ M±SD	F coef.	k M±SD	F coef.	3/k M±SD	F coef.	5/k M±SD	F coef.
Beech	0.402±0.017	3.657 ***	0.398±0.054	1.412 *	7.639±0.951	5.941 *	12.732±1.586	5.942 *
Douglas fir	0.199±0.033		0.289±0.064		10.792±2.319		17.987±3.865	

ANOVA, n=5, values are mean (S.D.), levels of significance: ***p<0.001, *p<0.05

depths of 0-10 cm (0.021±.002:0.067±0.007, p<0.001) and 10-20 cm (0.012±0.002:0.035±0.003, p<0.001). The content of easily accessible K in the Douglas fir culture soil was less only in the surface layer of 0-10 cm (0.25±0.024:0.30±0.029, p<0.01), while at depths of 10-20 cm (0.20±0.019:0.16±0.017, p<0.001) and 20-50 cm (0.065±0.006:0.050±0.004, p<0.001) it was higher than in the soil at the control stand. The C content (C g kg^{-1}) in the soil at both sites fell with depth of soil horizon (Table 2). The soil at the Douglas fir culture had a lower C content at all depths, except at 50-80 cm where no differences were observed.

The C/N ratio at both sites was highest in the litter layer and decreased with depth of horizon (Table 2). A higher C/N ratio was found in the leaf litter at the Douglas fir stand than at the control site (27.76±2.31:21.09±1.98, p<0.001), as was the case for the surface soil layer (14.09±1.22:11.82±0.96, p<0.001) and soil layer 10-20 cm (12.04±1.23:10.28±1.01, p<0.01). In the deeper layers of soil (20-50 cm and 50-80cm), the ratio became greater at the control beech stand than at the Douglas fir culture (p<0.001; p<0.01).

Litter decomposition

The amount of litter accumulated on the forest floor in the Douglas fir culture was significantly greater than that in the beech stand (12.791±4.021 t ha^{-1} :7.482±2.368 t ha^{-1}, p<0.001). A slower decomposition of Douglas fir organic matter in comparison to beech organic matter was observed throughout the experiment (Table 3). After six months, Douglas fir matter had decomposed 9.48±1.51% in comparison to 18.2±0.7% for beech matter (p<0.001). After a year, Douglas fir organic matter had decomposed 24.98±4.74%, as opposed to 32.76±3.57% for beech matter, (p<0.05). An analysis of the decomposition constants (k½ and k) and decomposition prognosis (3/k and 5/k) showed that at each stage of the experiment, the organic matter from Douglas fir decomposed more slowly than that of beech (Table 4).

The decomposition prognosis of the organic matter of the examined species, according to Olsen's decomposition model, showed that 95% of the Douglas fir leaf litter will take 10.729±2.319 years to decompose, while the same percentage of beach litter will take 7.639±0.951 years. Decomposition of 99% of the Douglas fir organic matter will take 17.987±3.865 years, and beech organic matter will take 12.732±1.564 (Fig. 4)

DISCUSSION

Floristic composition

Many studies have shown that the content, abundance and cover of understory species depends on the species of trees present in the overstory. Namely, plants in the herbaceous layer are affected by the tree layer, which modifies the transmittance of light, the water balance and microclimate, litter characteristics, chemical characteristics and porosity of the soil, as well as through leaching of toxic compounds [1,5,11,35,39,50]. In addition, silvicultural management, former land use and atmospheric deposition have significant impact [1].

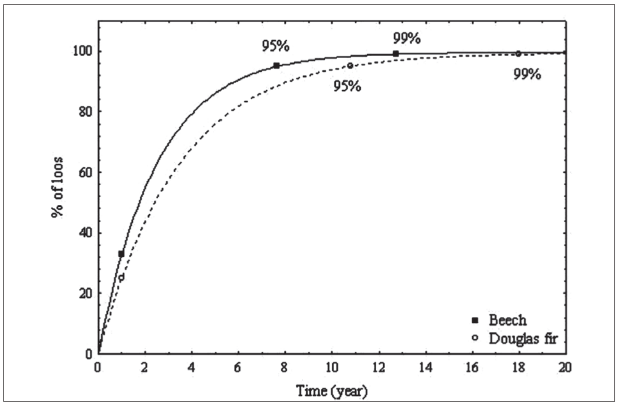

Fig. 4. Proposed model for the process of organic matter decomposition in beech and Douglas fir stands.

Through phytocoenological research, we found depletion in floristic content in the Douglas fir culture. The dense planting of Douglas fir and low-intensity thinning cuts have led to a modification of environmental conditions (reduction in the quantity and quality of light), which has caused the modification of the ground vegetation on this plot [5]. In the sciophilous Douglas fir culture, we observed a complete absence of shrub layer. In the herbaceous plant layer, the highest number and cover are those of *Galeobdolon luteum, Viola silvestris* and *Rubus hirtus*, which, along with *Cardamine bulbifera*, represent the most numerous and frequent herbaceous species in Douglas fir cultures in western Serbia. However, missing from the herbaceous layer are many of the plant species characteristic for natural beech forests, such as *Mycelis muralis, Asperula taurina, Allium ursinum, Helleborus odorus, Epilobium montanum, Circaea lutetiana, Glechoma hirsuta, Sanicula europaea* and many more. Similar results are found in the comparative phytocoenological research performed in the young beech forests and Douglas fir cultures in the region of Bukovo near Kosjerić in western Serbia [39]. We also observed the presence of young beech in the Douglas fir culture, which is a sign that artificial stands of conifers are rejuvenated by beech, i.e., succession moves toward autochthonous forest vegetation [39]. On the other hand, in a study at 12 localities in the Czech Republic, it was established that the cultivation of Douglas fir was reflected in an increase in diversity, but also in a decrease in the abundance of ground vegetation at the sites [35]. The impact of Douglas fir was most pronounced in comparison to managed Norway spruce stands. However, the above-described differences in understory are not so noticeable when European beech stands are substituted by Douglas fir. The results of our research indicate that with the application of more intensive thinning of the Douglas fir culture in a beech habitat, the ground flora does not differ significantly from that in deciduous forests, as has been shown in the research of other authors [1,51]. Nonetheless, the current state of the herbaceous cover in the Douglas fir culture, with its significantly reduced abundance and cover of understory species indicates that the planting of this species in a beech habitat correlates negatively to floristic composition, i.e. biodiversity of the habitat.

Litter and soil analysis

Many studies have found that the surface layer of soil in coniferous forests, particularly spruce stands, is drier than in deciduous forests. Namely, coniferous forests are characterized by a higher degree of interception and a longer period of transpiration [53]. On the contrary, the results of our research show no difference between the average moisture content in the surface layer soil of the Douglas fir culture and the beech stand during the vegetation period. The only difference was found at the depth of 50-80 cm, where higher moisture content was found in the Douglas fir culture. It is known that different structures of plant root systems and the depth of their spreading have significant impact on the moisture of the soil in which they grow. In addition to the strong heart root system with which it draws water from the deeper layers of soil, beech has a concentration of finer roots in the topmost soil layer, while the density of the fine root vessels of Douglas fir is constant in the deeper layers [1,33,54]. The vertical distribution of the fine vessels of a root system is best seen in mixed Douglas fir/European beech stands [55]. In addition, herbaceous plants, whose cover is 100% in the beech stand, have a thicker distribution of roots in the surface layer of soil, making water collection much more efficient [56]. The vertical distribution of the root systems of beech and Douglas fir, along with the marked difference in the viability of the herbaceous cover and differences in tree layer density and interception ability, affect the moisture levels in the soil horizons of the examined sites.

The substitution of deciduous woody species with fast-growing coniferous cultures most often brings about the acidification and depletion of the soil in forest ecosystems in moderate climate zones, which is generally characterized by an insufficient nutrient content [57]. The impact of woody plants on the acidification of soil includes modifications to the intensity of finding and storing nutrients, the intensity of interception of acid pollutants from the atmosphere, the concentration of anions in the soil and soil solutions, amount of organic acids and degree of acid protonation, as well as mineralization, nitrification and weathering rates [1,2,17,58]. Some authors consider the substitutional acidity of soil a much more precise indicator of the effect of some plant species on soil acidity than active acidity, while others emphasize that analysis of the adsorption complex, in particular the degree of saturation of the adsorption complex by base cations is more reliably assessed by examination of the progress of the acidification process [59,60]. Our research found no difference in active acidity values, while a more acidic pH_{KCl} ($p<0.05$) was measured in the upper soil layer (0-10 cm) under Douglas fir. A significant reduction was observed in all parameters of the adsorption complex ($p<0.001$) at depths of 0-10 cm and 10-20 cm (lower values of T, S and V), as well as a lower C content ($p<0.001$; $p<0.01$) in the Douglas fir stand compared to the beech stand (control). Most studies on the impact of tree species have demonstrated an acidification of soil under coniferous trees when compared with hardwoods, these changes being most pronounced in the surface layer of soil [4-6,15,16,61,62]. However, numerous analyses have shown that the acidification impact of Douglas fir is less than that of spruce [24,34,37,63]. No difference in active acidity (pH_{H2O}), but also a lower value of substitutional acidity (pH_{KCl}) in a Douglas fir culture compared to an adjacent broadleaved stand was described [34]. A pronounced accumulation of humus on the forest floor and lower levels of soil acidity in a Douglas fir stand was reported [63], as well as deterioration in soil properties with increased representation of Douglas fir in a mix with beech, primarily observed as a reduction of base cations (Ca and Mg) in the surface soil layer [64]. The organic matter content in the soil has a significant effect on both cation exchange capacity and soil pH, especially in the surface layers [2]. In many countries, Douglas fir is known as a very productive species, characterized by a higher accumulation of surface organic matter compared to broadleaved species [32,34,65]. However, its growth and organic C turnover are limited by different climatic conditions as well as the chemical characteristics of the organic matter [8]. Under conditions of continental climate, the C input in soil, mineralization rates and size of the labile C pool are reduced in Douglas fir cultures [66]. The Ca content in organic material improves the dynamics of soil fauna populations and the process of mineralization. The lower content of this element in Douglas fir litter compared to beach litter [8,34,64,67] contributes to a slower mineralization and more limited incorporation of the organic matter that accumulates on the surface soil in a the culture [1]. The lower C content in the soil that we found in the Douglas fir culture shows that the amount of incorporated organic matter is less than that in the soil of the beech stand, causing the reduced cation exchange capacity under Douglas fir. Nonetheless, the characteristics of the adsorption complex in the Douglas fir culture are better when compared to other conifers, e.g. spruce [2,8,62,65]. Due to the smaller cation exchange capacity under Douglas fir, the number of negative charges balanced by H^+ decreased and fewer protons were exchanged with the solution, resulting in less acid pH values. Similar results for the impact of Douglas fir on pH, cation exchange capacity and carbon content in soil were presented [2].

Based on our research, significantly lower levels of nitrogen ($p<0.001$) and available forms of phosphorus ($p<0.001$) and potassium ($p<0.01$; $p<0.001$) in the two surface soil layers at the Douglas fir stand were found compared to the beech stand (control). Since the maintenance of certain reserves of nutrients is a basic prerequisite for the stability of an ecosystem, the results of our research indicate that the substitution of the autochthonous beech stand with a Douglas fir culture has brought about a decrease in the stability of the beech habitat. Various studies have shown that the input-output budget of nutrients for hardwood stands is close to equilibrium, whereas conifer stands in the same location have a significantly negative balance [1,68,69]. This is a consequence of the higher concentration of nutrients in deciduous litter compared to conifer litter, as well as the fact that the loss of nutrients, through leaching, is greater in conifer stands than in beech [8,34,70,71]. One such study of the nutrient dynamics in chronosequence of Douglas fir stands in France revealed the negative input-output budgets for N, S, K, Ca and Mg, which depletes the soil [72]. The substitution of beech stands with a Douglas fir culture is reflected in the total P and K contents in the surface soil layers [8,70], with which our results are in agreement with. However, our results are incompatible with the observation that a change in vegetation does not have a significant effect on the total N content. One of the most important functions of soil is its ability to retain N because it has an important role in terms of plant nutrition [23]. Woody species, as proton donors (nitric and organic acids) at the forest floor level, have the potential to influence the evolution of pedogenetic processes in the lower mineral horizon [73]. Corresponding anions may induce mineral weathering and leaching of cations and metals from topsoil, which brings about nutrient depletion, soil acidification and podzolization [74-76]. In monoculture Douglas fir stands on acid soil the level of nitrification is increased, but the intensity of this process exceeds the immobilization of nitrates by microbe populations and their uptake by Douglas fir roots [77,78]. This inevitably brings about leaching losses of nitrates, which are always high in Douglas fir stands. The reduction of the N content in the soil of Douglas fir cultures could additional intensify the acidification of this habitat in the future. Results from our study showed that the largest contribution of nitrogen is concentrated in the surface (0-10 cm) soil layer in the beech stand where a greater presence of nitrophilic plants (*Urtica dioica, Allium ursinum, Stachys silvatica, Circaea lutetiana, Moehringia trinervia*) in the herbaceous and shrub (*Sambucus nigra*) layers was found. The lower potassium content in soil populated by conifers compared to soil populated by deciduous forest has also been confirmed by other authors [4,6,61]. In addition, the increase in soil acidity in the Douglas fir culture that could take place in the future could result in a further reduction in the content of available P [74]. Although the litter of Douglas fill has a relatively favorable content of nutritive material and a preliminarily relatively good degradation of the litter, at least compared to spruce, for this very productive species to grow, it needs a considerable amount of nutritive material, which could bring about future depletion of the soil and a reduction in the productivity of the stand. The decrease in content of available forms of P, K, Ca and Mg in the soil of a Douglas fir culture has been established [63]. Apart from this, a higher nutrient content in the surface horizon-layer of the soil at the control site is also a result of the dense ground cover of herbaceous plants, which is very involved in their cycling [62], as well as beech acting as a pump that recycles nutrients from deeper soil horizons through its deeper root system [79].

The content of N and the C/N ratio on the forest floor are important parameters for determining the impact of tree species on ecosystem functioning. The variability of these parameters is closely linked to changes in tree species composition [23]. The C/N ratio in litter is one of the indicators of the intensity of the organic matter decomposition process in the ecosystem, and a ratio less than 25 in the primary organic source indicates that there is no hindrance to the decomposition of the organic material [80]. Our research highlighted a reduction in the N content in the soil of the Douglas fir stand in relation to the control site, which leads to an increase in the C/N ratio in the culture. The higher C/N ratio found in Douglas fir litter when compared to that at the control site (27.76:21.09) was the first indicator of the future deceleration of organic matter decomposition in the Douglas fir stand.

Differences in the intensity of decomposition of organic matter of different woody species is conditioned by the different organic compositions of litter and heterogenous distribution of carbon and nitrogen resources, and by differences in the composition of the decomposer community and microclimatic conditions (soil moisture, pH, temperature and fertility) of the stands [80]. In our study, the slower process of organic matter decomposition in the Douglas fir culture in comparison to the control site, and thereby slower process of nutrient cycling as well, was confirmed by the litterbag experiment. Our research shows that the planting of Douglas fir has caused changes in the microenvironmental conditions (reduction in quality and quantity of light and nutrients in the soil), which has led to the deceleration of the decomposition of organic matter in the Douglas fir stand. In addition, litter quality, i.e. its chemical composition, influences the decomposition process, so that in the early stage of decomposition (4-6 months) it is closely tied to the litter chemistry of water soluble nutrients and structural carbohydrates, while in the later stage (over 12 months) it is more linked to lignin content in the litter material [81-83]. Previous research has shown that Douglas fir litter decomposed more slowly than beech litter due to less favorable microclimatic conditions, lower contents of N, K and soluble compounds, and higher cellulose and hemicellulose contents, while differences in lignin content were not so pronounced (17.1% and 16.2% in Douglas fir and beech litter, respectively) [84]. Accordingly, the results of our research revealed that the greatest differences in the intensity of decomposition of litter of the examined tree species were evident six months after the start of the experiment when the Douglas fir litter decomposed more slowly than beech litter ($p<0.001$). After one year, although these differences in litter decomposition intensity were still present, a tendency toward a diminution of the difference was observed ($p<0.05$), which corroborates the established dissimilarity in the chemical content of Douglas fir and beech litter [84].

The accumulation of undecomposed organic matter and the sequestration of nutrients, due to the slower decomposition of Douglas fir needles, resulted in a reduction in the content of essential elements in the culture soil, i.e. a decrease in its fertility. The lower levels of some essential nutrients at the Douglas fir stand in comparison to the control site can lead to further deceleration of the decomposition process [7,85]. The decomposition prognosis for the organic matter of Douglas fir compared to beech (10.8:7.6 years for 95% of decomposition and 18:12.7 years for 99%, respectively) points to the fact that nutrients in the Douglas fir stand will be excluded from the cycling process for a longer period of time. The slower decomposition of organic matter in the Douglas fir culture gives rise to a greater production of organic acids and further acidification of the soil due to a reduction in the basic cation content in the soil [61, 86]. It should be emphasized that the exceptionally high requirement of the fast-growing Douglas fir for nutrients can also contribute toward a reduction in the nutrient content in the soil. All of this points to a continuation of the degradation process and decrease in productivity of this habitat.

CONCLUSIONS

The results of our research show that the right choice of woody species for the formation of forests is of crucial importance to sustaining the diversity, fertility and the ecological stability of a forest ecosystem. Namely, we highlight that the substitution of an autochthonous beech forest with a Douglas fir culture, contributed to the initiation of degradation processes in natural deciduous habitats on Mt. Maljen. Forty years after the planting of a Douglas fir culture, the absence of many of the herbaceous plants characteristic to beech habitats, caused a reduction in biodiversity. In short, the obtained results indicate that the habitats in which beech grows at its optimum are not suitable for the planting of conifers, including Douglas fir, suggesting that tree species are the drivers of important soil properties (acidity, organic matter trend), which in turn affect forest ecosystem functioning. Therefore, it is essential to evaluate by the use of models the impact that a species could have on the ecosystem functioning, or an advantageous and eco- sustainable cultivation of a forest species.

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